Introduction

Stem cells and their unique properties: Stem cells are special cells which not only have the ability of self-renewal but can also be a lifelong source of specialised functional cells of different human organs. Development of a human embryo into a healthy new-born child is possible because of the unique ability of embryonic stem cells to form different tissues and organs. Most adult human tissues and organs also have stem cells that can produce their functional specialised cells as and when required. The self-renewal ability of stem cells ensures that stem cells are not depleted and enough stem cells remain to produce sufficient number of specialized cells of that organ during the long human lifespan, until aging starts affecting stem cells.

Stem cells in Regenerative Medicine and human diseases: When a disease or injury causes severe depletion of the functional cells of a human organ or system, the function of that organ or organ system is lost. In the natural healing process, some organs such as skin, blood, liver etc. can often regenerate its form and function by producing sufficient numbers of new functional cells from the stem cells present in them. However, specialized cells of some organs like the nerve cells in the brain, spinal cord, eyes and muscles have limited or no capacity to regenerate and restore full function. In the last two decades, medical science has undertaken extensive research to explore the potential of stem cells from the same organ or tissue type (homologous use) or from a different organ or tissue type (non-homologous use) to restore some lost bodily function. These stem cells may be from the same person (autologous source) or from another person (allogeneic source). Research to regenerate the form and function of a human organ or organ system from stem cells or tissue engineering is called 'Regenerative Medicine'.

Status of Stem cells in Regenerative Medicine and human diseases: Unfortunately, the promise of Regenerative Medicine in general, and stem cells in particular, is yet to be realized due to several technical, biological, ethical and medical challenges. To produce sufficient number of specialised cells for restoring a lost body function with just a small number of stem cells or by using stem cells from one organ to restore cells and function of a different organ (such as mesenchymal stem cells in bone marrow or fat tissue to restore nerve or muscle function) has proven to be far more difficult in humans than what was thought based on animal experiments. As a result, the inherent appeal of stem cells has remained largely unfulfilled in human diseases. The exception is however the use of "Haematopoietic Stem Cells" for reconstituting or regenerating the bone marrow in order to start producing blood and immune

cells. Transplantation of enough number of "Haematopoietic Stem Cell" in a procedure called Bone Marrow Transplantation or Haematopoietic Stem Cell Transplantation from the same person (autologous) or from another human donor (allogenic) is a recognized medical indication of stem cell use for benign and malignant life threatening haemato-lymphoid diseases or few immune related diseases. Haematopoetic stem cells are also progenitors for other cells like osteoclasts and have successfully used in osteopetrosis and some inborn errors of metabolism like Gaucher disease, mucopolysaccharidosis. Use of other types of stem cells and even the bone marrow derived stem cells to restore function of other organs remains experimental and is subject of ongoing controlled clinical trials. Not only the efficacy of these experimental stem cell use is uncertain, the process of taking out stem cells, culturing or growing them, storing them and putting them back can cause changes in these cells and sometimes serious side effects, including some reported cases of cancers.

Why Stem cells continue to be used for debilitating or incurable conditions outside controlled research studies: A large number of controlled prospective research studies (phase I, II and III clinical trials) investigating the safety and efficacy of stem cells for different diseases have been completed or are ongoing in Europe, USA, Korea and Japan. A small number of such research studies are also being conducted in other countries, including India. All developed countries have taken a very cautious and stringent regulatory approach regarding how different types of stem cells can be procured, processed, stored and used for preclinical or clinical research or as stem cell therapy outside research studies. Participants of regulated interventional research in any field, including stem cells, are made aware through a detailed written informed consent process about the experimental nature of the therapy, unproven efficacy and uncertainty regarding the benefits and risks of stem cells, the natural history of the disease, current standard therapy for that disease and any alternative treatments. It is the duty of the research sponsors to provide free of cost medical tests and treatments done as part of stem cell clinical trial and research, including the cost of procuring, storing and using stem cells. Circumventing the route of rigorous research studies to establish the safety and efficacy of a particular type of stem cells for a specific disease or aging condition, some unlicensed or even licenced and registered medical practitioners engage in unethical practices of selling unproven stem cell therapy as a magical remedy to desperate families with incurable and potentially fatal diseases with little or no hope of cure from other methods. Desperate patients from around the world including USA and Europe with stricter enforcement of regulations for stem cell use outside clinical trials get lured to stem cell clinics in South America, China, Russia and India. The US FDA and European Medical Agency has warned against this practice through several such advisories.

https://www.fda.gov/consumers/consumer-updates/fda-warns-about-stem-cell-therapies

https://www.fda.gov/news-events/press-announcements/statement-stem-cell-clinic-permanent-injunction-and-fdas-ongoing-efforts-protect-patients-risks

https://www.fda.gov/news-events/press-announcements/federal-court-issues-decision-holding-us-stem-cell-clinics-and-owner-adulterated-and-misbranded-stem

Is Stem cell research permitted or encouraged by the governmental agencies?

The unethical and unregulated use of stem cells as, often promoted as a magical remedy is not allowed by the government in the developed world and many Low and Middle Income Countries (LMIC) including India. However, considering the incurable nature of many diseases, and the acknowledged potential of stem cells, most countries, including India, encourage and fund scientific, ethical and regulated research in the field of stem cells. The purpose of such research is to obtain safety and efficacy data with the use of a particular type of stem cell in a particular condition. To provide guidance and to facilitate human research in stem cells, while curbing exploitation of vulnerable patients, the Indian government through the Indian Council of Medical Research (ICMR) has come out with successive National Guidelines in this field since 2007. The most recent National Guidelines for Stem Cell Research with inputs from all stakeholders including various government agencies and regulators, patients, medical and scientific experts and the industry, was released in 2017. These guidelines are revised at regular intervals to incorporate any new evidence for the safety or efficacy of stem cells.
https://www.icmr.nic.in/sites/default/files/guidelines/Guidelines_for_stem_cell_research_2017.pdf

Need for National Guidelines for evidence-based use of Stem cells as a routine or standard treatment option: In many countries including India, there is a lack of clarity among patients, and to some extent among the medical community, whether stem cell therapy can be considered as a standard treatment option for a specific medical condition or should remain as an unproven experimental approach. There are several reports of increasing use of stem cells therapy for a wide range of diseases, often with little or no scientific evidence of efficacy or cure. Unethical promotions with false claims and misleading advertisements have been widely used to promote unscientific stem cell therapy. Several instances of public exploitation and grievances from members of the public have been received by the ICMR and other government agencies from aggrieved patients describing how they were lured into unproven stem cell therapies. Often the complainants demanded actions to be taken by the regulatory agencies and professional bodies to curb such practices. With this background, the Govt. of India has entrusted the ICMR to frame guidelines on stem cell therapy.

In order to develop a scientific and unbiased guideline for evidence based use of stem cell as a routine or standard treatment option in India, the ICMR has solicited opinion from expert clinicians, professional medical societies and through its website from any clinician or member of public to submit level I or level II scientific evidence for clinical efficacy of stem cells in any disease indications with reference for such evidence from peer reviewed Pubmed indexed medical and scientific journals.

https://icmr.nic.in/content/icmr-inviting-level-i-or-level-ii-scientific-evidence-and-grade-or-b-recommendation-use-stem

A critical review of the comments and evidence provided by medical experts and their professional societies or any member of the public and the scientific literature was done to draft guidelines and statements for evidence-based use of stem cell therapy.

Statements have been prepared for individual diseases or groups of diseases or conditions on the "EVIDENCE BASED STATUS FOR THE USE OF STEM CELLS IN (Disease condition)". In these statements the first section is for the public and patients using layman terms while the second section is for doctors, scientists and allied healthcare professionals providing major research studies in the scientific literature, scientific level of evidence and a summary recommendation based on the current scientific evidence.

International Society for Stem Cell Research (ISSCR)

The International Society for Stem Cell Research (https://www.isscr.org/) is the leading professional organization of stem cell scientists and represents over 4,000 members in 67 countries including India. Like ICMR in India, FDA in USA, EMA in Europe, this international society also felt the urgent need to address the growing public concern regarding the unscientific or unethical use of stem cell therapy. The ISSCR has also issued a statement on reporting false marketing claims and adverse events from clinics offering unapproved stem cell therapies.

https://www.closerlookatstemcells.org/patient-resources/how-to-report-false-marketing-claims-and-adverse-events-from-clinics-offering-unapproved-stem-cell-therapies/.

In parallel with the ICMR initiative and public advertisement inviting comments and evidence for stem cell use from public and medical professionals, the ISSCR has also come out with factsheets on current status of stem cell use. The ISSCR document highlights that other than Hematopoietic stem cell (also called Bone Marrow) transplant for certain haematological or immune system disorder, the "list of diseases for which stem cell treatments have been proven

to be beneficial and/or have obtained regulatory approval for use is still very short" and that "some bone, skin and corneal (eye) injuries and diseases can be treated by grafting or implanting tissues in which stem cells are essential for the healing process".The ISSCR cautions that "However, clinics around the world continue to provide unproven stem cell treatments and often market them as cures for a variety of diseases and conditions without sound scientific evidence or regulatory approval. These so-called treatments have, in some cases, caused patients great harm physically, and at great expense financially".
https://www.isscr.org/professional-resources/scientific-professional-resources/disease-fact-sheets
https://www.isscr.org/scientific-clinical-resources/disease-fact-sheetshttps://www.closerlookatstemcells.org/2020/01/14/truths-around-stem-cell-treatments/

The ISSCR concise factsheets provide the current state of stem cell science for specific diseases, including background on the disease, rationale for using cell-based therapies, evidence for specific approaches and current status of the field with respect to clinical trials. A total of 11 conditions have been covered so far.

1. Age-related macular degeneration
2. Amyotrophic lateral sclerosis
3. Chronic obstructive pulmonary disease
4. Diabetes
5. Huntington's disease
6. Liver disease
7. Multiple sclerosis
8. Myocardial infarction / Heart failure
9. Osteoarthritis
10. Parkinson's disease
11. Paediatric leukodystrophies

Evidence Based Status of Therapeutic Use of Stem Cells in Autism Spectrum Disorder (ASD)

A. Information for public and patients

What is Autism or Autism Spectrum Disorder?

It is a neurological and developmental disorder which is first noticed in childhood and the condition remains the same or worsens in later life. It is termed "Autism Spectrum Disorder" as may cause a range of problems from difficulties in speaking, avoiding eye contact and doing or saying things repetitively. If ASD is suspected in your child, a more detailed evaluation is done by a team of specialists to make a diagnosis of ASD. Some genetic disorders namely, fragile X syndrome, Rett syndrome, Tuberous sclerosis have autistic features. However, there are no known or well-established causes for most of the cases of ASD.

What is the treatment of autism?

Patients with autism are advised behavioural and occupational therapy along with other forms of supportive therapies as required. Sometimes pharmacological agents are used for symptomatic management of certain manifestations of Autism spectrum disorder.

Have stem cells been used in ASD?

Along with supportive therapies and drug treatment, few studies have tested the use of various forms of stem cells to improve the outcome in children with Autism. We are aware that many Indian patients with ASD have been offered different types of stem cell therapies as a standard treatment option and not as part of any approved clinical trial / research. ICMR with inputs from medical specialists in this field has reviewed the existing scientific and medical literature and submissions from practicing doctors and their professional societies regarding any evidence-based safety and efficacy of stem cells in ASD. ***Critical review of the studies reported so far does not support the use of stem cell therapy over and above the behavioural and supportive therapies for ASD.***

Recommendations (2021): Based on the review of available scientific evidence, stem cell therapy should NOT be offered as a standard or routine therapy to patients with Autism. These guidelines will be periodically reviewed for any new evidence showing benefit or harm with the use of stem cells for Autism Spectrum Disorder. Therapeutic use of any type of stem cell in Autism should be restricted to clinical trials only after obtaining necessary regulatory

approval as defined in National Guidelines for Stem Cell Research-2017. The patients participating in these clinical trials should be closely monitored for the possibility of any harm with use of stem cells. As per the ICMR National Bioethics Guidelines 2017, trial participants should have read and signed the informed consent form which explains them the alternative therapies, possible benefits as well as harm due to experimental treatments like stem cell therapy. Participants should not be made to pay for any expenses incurred beyond routine clinical care and which are research related including tests, investigations and any interventions (such as stem cells). This is applicable to all participants, including those in comparator/control groups. Participants in a clinical trial should be provided compensation in the event of any harm or permanent injury or death due to the use of experimental stem cell therapy.

B. Information for Medical / Scientific / Allied Health Professional

The diagnosis of Autism Spectrum Disorder is made after a detailed evaluation by a multidisciplinary team. There are very few established causes of ASD and this remains an active area of research. Based on their symptoms and signs, ASD patients are managed with behavioural and occupational therapy along with other forms of supportive therapies. In some ASD cases, pharmacological agents are used in an attempt to control some manifestations. Along with these supportive therapies and drug treatment, there are several reports of the use of stem cells to children with Autism. Many Indian patients with ASD have also been offered different types of stem cell therapies as part of research studies and also as a standard treatment option which is outside the purview of approved clinical trial. ICMR with inputs from experts in this field has reviewed the existing scientific and medical literature and submissions from practicing doctors and their professional societies regarding level of evidence for efficacy and safety of stem cells in ASD. A critical review of the published human studies that are either randomized controlled trials or have been submitted in response to the ICMR call for Level I/II evidence supporting the use of stem cells in Autism Spectrum Disorder has been undertaken. Summary of some representative studies is outlined below:

Autism Spectrum Disorder	
S.No.	**Review of Literature** **Critique / Applicability of the study results**
i.	Source:Human embryonic stem cell RoA:IV, IM, Epidural, popliteal block, brachial plexus block, intrathecal, epidural catheter caudal, deep spinal muscle Shroff G. Human Embryonic Stem Cells in the treatment of Autism: A case series. InnovClin Neurosciences. 2017, 14 (3-4), 12-16 PMID: 28584692 Three cases studied. No control used. The patients were treated with 3 to 4 sessions from 2011 to 2013. 'Results: The patients showed improvements in eye coordination, writing, balancing, cognition, and speech and showed reduced hypersensitivity to noises and smells.' The time period of post-treatment evaluation has not been given. The patients were given occupational and physical therapy additionally. It is mentioned that there was improvement but tools for assessing behavioural & intellectual functions have not been described and there is no objective assessment pre- and post-therapy. The paper is published in 2017, but there is no data about follow up after therapies given in 2012-13. The weakness in this study prevents its findings from being used to support the use of stem cells.
ii.	Source:Human cord blood mononuclear cells (CBMNCs) & umbilical cord derived mesenchymal stem cells (UCMSCs) RoA:Combined IV and IT transplantation Lv YT, Zhang Y, Liu M, Qiuwaxi JN, et al. Transplantation of human cord blood mononuclear cells and umbilical cord-derived mesenchymal stem cells in autism. J Transl Med. 2013;11:196. doi: 10.1186/1479-5876-11-196. This non-randomized, open-label, single center phase I/II trial from China showed that combined (CBMNC and UCMSC) therapy showed significant improvement in CARS score by 24 weeks. However, it is a small study of 37 cases. It was an open label and non-randomized study.After the 2013 publication, there has been no follow up publication to see if there was any long-term benefit or harm
iii.	Source:Autologous umbilical cord blood (AUCB) RoA: Peripheral IV infusion Chez M, Lepage C, Parise C, et al. Safety & Observations from a Placebo-Controlled, Crossover Study to Assess Use of Autologous Umbilical Cord Blood Stem Cells to Improve Symptoms in Children with Autism. Stem Cells Transl Med. 7(4):333-341 A randomized, blinded, placebo-controlled trial but in only 29 children with autism. With autologous umbilical cord blood infused intravenously a trend towards improvement, particularly in socialization, was seen but there was no statistically significant differences for any endpoints

iv.	Source:Fetal stem cells RoA:IV or subcutaneous Bradstreet JJ, Sych N, Antonucci N, et al Efficacy of fetal stem cell transplantation in autism spectrum disorders: an open-labeled pilot study. Cell Transplant. 2014;23Suppl 1:S105-12. doi:10.3727/096368914X684916 This study used Fetal stem cells and relied on Autism Treatment Evaluation Checklist (ATEC) Structure to judge efficacy. ATEC is a caregiver-administered questionnaire designed to measure changes in severity of ASD after treatment. Caregiver's assessment is likely to be a source of bias. Also the ATEC scores decrease with increasing age and this has not accounted for while interpreting the decrease in score as the efficacy of stem cell treatment. (See study on ATEC -Mahapatra S, Vyshedsky D, Martinez S, Kannel B, Braverman J, Edelson SM, Vyshedskiy A. Autism Treatment Evaluation Checklist (ATEC) Norms: A "Growth Chart" for ATEC Score Changes as a Function of Age. Children (Basel). 2018. 16;5(2). pii: E25. doi: 10.3390/children5020025)
v.	Source:Autologous bone marrow mononuclear cells RoA: Intrathecal Sharma A, Gokulchandran N, Sane H et al. Autologous bone marrow mononuclear cell therapy for autism: an open label proof of concept study. Stem Cells Int. 2013;2013:623875. doi: 10.1155/2013/623875. In a small study of 32 patients with Autism, autologous bone marrow mononuclear cells (BMMNCs) were given intrathecally along with multidisciplinary therapies. A significant improvement in certain scores was reported at a mean follow up of 12 months. Under the heading limitations and future directions, the authors acknowledge that *"The study is an open label proof of concept. A small sample size, the absence of randomization, and the absence of control group were the limitations. Large scale, multicentre, and randomized controlled trials are recommended. A longer period of follow up may be required to further establish the safety and efficacy. Few patients had increased episodes of seizures after the intervention, which were controlled with medications…".* In fact, the 9% seizure rate after this therapy is a matter of concern. Without any follow up randomized or larger Phase II study after the 'Proof of Concept' study of 2013, its findings cannot be used to justify stem cells as a standard therapy option.
vi.	Source:Autologous umbilical cord blood RoA:IV Dawson G, Sun JM, Davlantis KS, et al. Autologous Cord Blood Infusions are Safe and Feasible in Young Children with Autism Spectrum Disorder: Results of a Single-Center Phase I Open-Label Trial. Stem Cells Transl Med. 2017; 6(5):1332-1339 Pre and post assessments were done based on caregiver and clinician administered tools. Out of multiple scales used, most did not show significant p values. Discussion mentions about improvement in scores but does not refer to the p value for statistical significance.

vii.	Source:Autologous bone marrow aspirate concentrate RoA: Intrathecal Bansal H, Verma P, Agrawal A et al. A Short Study Report on Bone Marrow Aspirate Concentrate Cell Therapy in Ten South Asian Indian Patients with Autism. J Stem Cells. 2016;11:25-36 In this study, intrathecal transplantation of bone marrow aspirate concentrate stem cells was performed. The maximal effect of cell therapy was observed within the first 12 months following the treatment. Interestingly, they also found that improvement decreased with increasing age of the child. The authors acknowledge it was a pilot study and its findings cannot be used to justify stem cells as a standard therapy option.
viii.	Sharifzadeh N, Ghasemi A, TavakolAfshari J, Moharari F, Soltanifar A, Talaei A, Pouryousof HR, Nahidi M, FayyaziBordbar MR, Ziaee M. Intrathecal autologous bone marrow stem cell therapy in children with autism: A randomized controlled trial. Asia Pac Psychiatry. 2020 Nov 4:e12445. doi: 10.1111/appy.12445. Epub ahead of print. PMID: 33150703. Overall, 32 patients in two groups of intervention (n = 14) and control (n = 18) completed the study, of which 27 (84.4%) were male. Mean age was 9.50 ± 2.14 years. The improvements in CARS total score, GARS-II autism index, and CGI global improvement showed **no significant differences between the groups over 12 months.**
ix.	Price J. Cell therapy approaches to autism: a review of clinical trial data. Mol Autism. 2020 May 24;11(1):37. doi: 10.1186/s13229-020-00348-z. PMID: 32448347; PMCID: PMC7245880. The author is from the institute of psychiatry, Kings College, London. In this article all publications and clinical trials are reviewed. The author concludes that, these studies present a mixed picture. The only placebo-controlled study resulted in a negative outcome, while the open-labelled studies provided mixed and, in most cases ambiguous, outcomes. The author gives the following conclusions and raises a strong question about exposing the children to probably unsafe and ineffective therapies. "A number of reservations arise from this tranche of studies, specifically the absence of identified therapeutic targets, and deficiencies in the therapeutic approach that is being employed. " "The data on advanced therapies is currently too sparse to analyse robustly, but the experimental nature of these therapies means that their success rate is unlikely to be higher. This means that the overwhelming majority of patients taking part in trials such as those considered here are receiving treatments that are unsafe, ineffective, or both. Parents and clinicians would do well to remember that these patients, for the most part, are children, unable themselves to give consent. In many cases, the future quality of life is very difficult to assess. How legitimate is it to expose these individuals to risk with such a low probability of success?

Unfortunately, most of these studies have one or more flaws which prevent us from drawing an unbiased and valid conclusion to support the use of stem cell therapy over and above the behavioural and supportive therapies. These studies are not double-blind randomized studies and have other shortcomings like small number of patients, not using unbiased tools for objective quantification of pre- and post-stem cell therapy cognitive function and behaviour,

absence of long term follow up data and relying on caregivers assessment for benefit with stem cell therapy. As these studies have used behavioural therapy along with the stem cell therapy it is not clear if the observed benefit is from intensive supportive therapy during the initial period and at long term follow up.

Summary of Evidence and Recommendations for Medical / Scientific Professionals (2021)

Based on the review of available scientific evidence, stem cell therapy should NOT be offered as a standard or routine therapy to patients with Autism Spectrum Disorder.

CAUTIONARY NOTE

The experts observed that severe autism can have a major impact on the quality of life of the affected child and the family. There is therefore a need to undertake research into the causes and more effective management of ASD. Since conventional management fails to control symptoms in many cases, such families see hope in some miraculous recovery with the use of stem cells without understanding the risks versus benefit ratio. It is therefore imperative that use of any type of stem cells in ASD should be restricted to clinical trials with necessary approval from regulatory authorities in India and as per the National Guidelines on Stem Cell Research - 2017.

https://www.icmr.nic.in/sites/default/files/guidelines/Guidelines_for_stem_cell_research_2017.pdfAs per the ICMR National Bioethics guidelines 2017 https://www.icmr.nic.in/sites/default/files/guidelines/ICMR_Ethical_Guidelines_2017.pdf,trial participants should have read and signed the informed consent form which explains them alternative therapies, possible benefits as well as harm due to experimental treatments like stem cell therapy. Participants should not be made to pay for any expenses incurred beyond routine clinical care and which are research related including tests, investigations and any interventions (such as stem cells). This is applicable to all participants, including those in comparator/control groups. Participants in a clinical trial should be provided compensation in the event of any harm or permanent injury or death due to the use of experimental stem cell therapy.

These guidelines will be periodically reviewed for any new evidence showing benefit or harm with the use of stem cells for Autism Spectrum Disorder.